

Larva and pupa of *Amyna axis* (Guenée, 1852) and affirmation of its taxonomic placement in Bagisarinae (Lepidoptera, Noctuidae)

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Abstract

The larva and pupa of *Amyna axis* (Guenée, 1852) are described and illustrated, and observations are provided on the insect's life history and larval biology. Larval, adult, and life history characters support the transfer of *Amyna* Guenée from Acontiinae Guenée, 1841 to Bagisarinae Crumb, 1956. The phylogenetic placement of the Bagisarinae is enigmatic; some adult and larval features indicate that the subfamily is a basal trifold proximate to Acontiinae, whereas other larval and life history characters suggest an association with Scoliopteryginae, a basal quadrifold group. Larvae exhibit a green-to-black color polyphenism presumably linked to larval density, with darker phenotypes occurring during outbreak densities. Parallel color shifts that occur in other Lepidoptera are briefly discussed.

Keywords

Larval polyphenism, larval melanism, thrashing behavior, frass flicking, Acontiinae, *Anomis*, *Amyna axis*, Bagisarinae

Introduction

The genus *Amyna* Guenée, 1852, contains about 35 recognized species distributed worldwide (Savelle 2009). The majority of the species occur in Asia. Only three are known from the New World; the pan-tropical *Amyna axis* Guenée, 1852 [= *A. octo*

(Guenée, 1852)], *A. bullula* (Grote, 1873), and *A. amplificans* (Walker, 1858). Forbes (1954) placed *Amyrna* in his concept of the Acontiinae, but noted that it was “A curious genus, probably not really Acontiine though with proper M of hindwing....” He also mentioned the presence of a blunt appendiculate tooth on the larval crochets (Fig. 7) that suggested the genus was related to “the Malvaceous group” of his Acontiine [*Bagisara* presumably]. Franclemont and Todd (1983) left *Amyrna* in the Acontiinae, grouping it in their concept of the Eustrotiini, a heterogeneous assemblage of taxa that includes genera now classified in the Acontiinae, Acronictinae, Condicinae, Eubleminae, Plusiinae, Xyleninae, etc. (Kitching and Rawlins 1998, Fibiger and Lafontaine 2005, Lafontaine and Schmidt (in press). Poole (1989) left *Amyrna* in the Acontiinae. Kitching and Rawlins (1998) mentioned *Amyrna* in their synopses of both Eustrotiinae and Bagisarinae, noting that the studies of Jeremy Holloway indicated the genus was better placed in the latter subfamily. Holloway (2009) tentatively moved the genus into Bagisarinae in his latest volume on the Moths of Borneo.

Amyrna axis, the type-species for Guenée’s genus, is the focus of this paper and the basis for the placement of the genus in Bagisarinae. Below we describe the larva and pupa, extending the efforts of Gardner (1941, 1946) and provide information on *Amyrna*’s larval biology. We discuss morphological, behavioral, and biological data that support the taxonomic assignment of *Amyrna* in Bagisarinae, and conclude the paper with a discussion of the green-to-black color polyphenism that occurs in *Amyrna* and other Lepidoptera.

Materials examined and methods

Larvae of *Amyrna axis* were obtained from *Amaranthus* L. in Scissors, Hidalgo County Texas in October 2006 (DLW) and October 2009 (Berry Nall), and *Croton* L. in Starr County, Texas in October 2008 (DLW and Henning von Schmeling).

One larva from Scissors was prepared for SEM study by running it through a series of ethanol baths (70%, 80%, 90%, 95%, 100%) before it was dehydrated with hexamethyldisilazane. The caterpillar was then coated with gold palladium for three minutes in a Polaron E 5100 sputter coater. Images were obtained with a Zeiss DSM-982 Gemini FE SEM at 3 kV. Larval, pupal, adult, film, and digital vouchers have been deposited at the University of Connecticut.

Descriptions

Description of living last instar (Figs 8–10, 12). Elongate, approximately 10 × longer than wide, shallowly constricted between segments, especially rearward. Ground color glaucous green, sea green, or emerald green to nearly black, often with black supraspiracular spots. Thin, white, broken addorsal, subdorsal, and supraspiracular stripes run length of body. Spiracular stripe continuous and roughly 2 × thickness of others,

weakening rearward of A8. Caudal segments somewhat flattened with anal prolegs elongated, splayed outward and held nearly horizontal behind body. Head shiny, pale green to orange, with setae borne from dark spots.

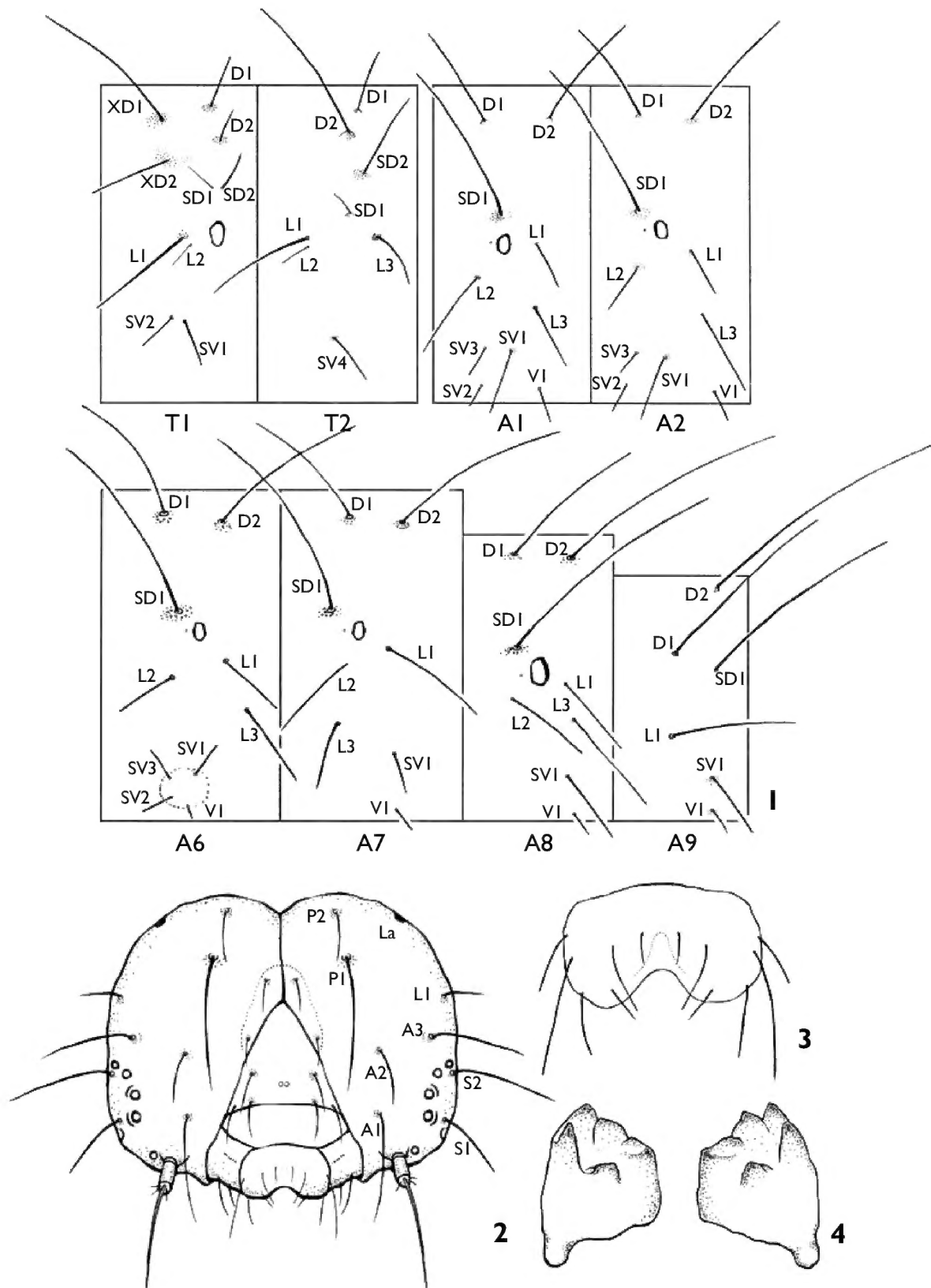
Description of preserved last instar (Figs 1–4). Length: 27–30 mm (n=15), body tapered caudad. Essentially unpigmented except for dark areas in melanistic individuals and darkened spots about some pinacula. Setae long, often blackened, frequently broken. **Head** – Primary setae above level of stemmata from blackened spots. P1 darkened, thick, extremely long, curving forward, reaching to clypeus; A3 darkened, thick, long, $>2 \times$ length of L1. La from darkened spot. Second segment of labial palpus long, $>5 \times$ basal diameter. Spinneret long and slender with apical pore (Fig. 6). **Thorax** – Prothorax with well-developed cervical gland (Fig. 6); XD1 long, reaching well forward; SD1 and SD2 approximate, at same level, SD1 fine, circa $1/5$ diameter of SD2; L1 second longest seta on segment reaching to stemmata; L2 fine, approximately $1/2$ length and $1/4$ diameter of L1; SV setae at same level, SV1 $2 \times$ length of SV2. T2 and T3 with D1, D2, SD2, and SD1, well separated, nearly in line; D2 seta longest; SD1 fine; L1 and L2 insertions approximate, but not sharing same pinaculum. **Abdomen** – A1–A9 with long setae; D2 and SD1 subequal to segment that bears them; D1, and especially D2, on raised warts that increase in size caudad to A9; D2 and SD1 on A9 longest setae on body, both exceeding length of A8 and A9; SD2 minute. L3 on A7 shifted anteriorad, below L2. A1 and A2 with three SV setae; SV1 $3 \times$ SV2 and SV3 on both segments; SV2 distant, shifted ventrad on A1, nearly in line with V seta. Spiracle height on A1 $1.2 \times$ than those on A2–A7; that on A8 $>2 \times$ that on A7; spiracles on A2–A4 shifted dorsad. Prolegs on A3 and A4 reduced to minute nib; those on A5 and A6 elongate, with 19–21 crochets. Crochets with blunt appendicular tooth (Fig. 7). A7–A9 with 3, 3, and 1 L setae, respectively. Anal plate with long setae; SD1 thickened and elongate.

Pupa (Figs 15–17). Length: 11.3–13.0 mm (n=3). Color as in Fig. 15. Deep incisures bounding two fully articulating segments A5–A6. Smooth except for darkened band of shallow pitting anterior to spiracles on A5–A7. Head structures poorly differentiated; only labrum and antennae fully delimited; labial palpus not visible. Prothoracic leg ending at about level of spiracle on A2.

Proboscis, meso- and metatarsi, and antennae all ending together between wing tips. Antennal segmentation vague. Small black pimplelike spot anterodorsal to spiracle on A5–A7. Cremaster consisting of single pair of spines, each shallowly hooked and slightly sinuate at apex.

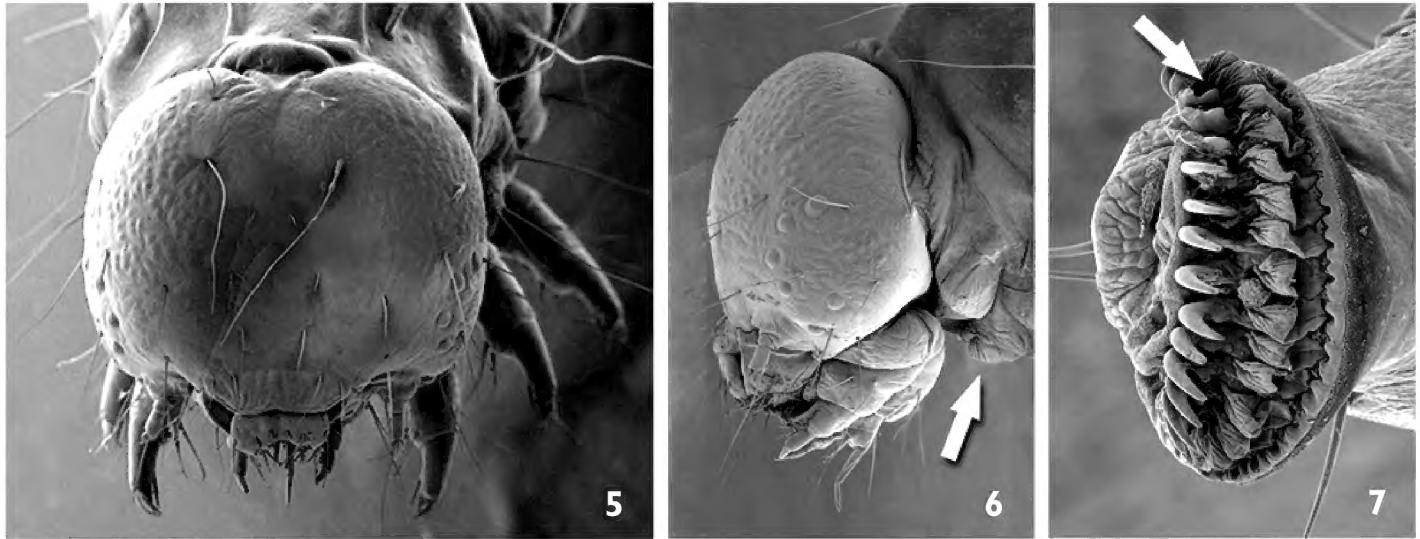
Life history

Host records include *Amaranthus* L., *Celosia* L., and *Digera* Forssk. (all Amaranthaceae); *Helianthus* L. (Asteraceae); *Chenopodium* L. and *Spinacia* L. (both Chenopodiaceae); *Ipomoea* L. (Convolvulaceae), *Ricinus* L. (Euphorbiaceae); *Arachis* L., *Crotalaria* L., *Medicago* L., *Phaseolus* L., and *Vigna* Savi (all Fabiaceae); *Hibiscus* L. (Malvaceae); *Car-*



Figures 1–4. *Amyna axis* last instar. **1** Chaetotaxy. SD2 on abdomen represented only by its pinaculum (forward of spiracle) **2** Head, frontal **3** Labrum, frontal **4** Mandibles, mesal surfaces.

diospermum L. (Sapindaceae); *Solanum* L. (Solanaceae); *Corchorus* L. (Tiliaceae); and *Parasponia* Miq. (Cannabaceae) (Ferguson et al. 1991, Robinson et al. 2001, 2002). Our southern Texas collections came from *Amaranthus* and *Croton* L., but not from any of the dozen or more other forbs and low-growing plants (e.g., *Abutilon* Mill., *Ambrosia* L., *Eupatorium* L., *Helianthus* L., *Heterotheca* Cass., *Lantana* L., *Malvastrum* A. Gray, *Melolochia* L., *Parthenium* L., *Physalis* L., *Sida* L., etc.) sampled with a beating sheet over the course of general (larval) collecting efforts in October 2006, 2007, and 2008. Nearly all of our *Amyna* caterpillars were collected from mature amaranth



Figures 5–7. *Amyna axis* last instar. **5** Head, frontal **6** Head, lateral, with prothoracic gland (adenosma) indicated **7** Crochets, with subapical tooth indicated.

plants, especially those that were into seed-set, turning red, and senescing. None came from lush, actively growing pre-flowering plants. *Amyna* caterpillars were so abundant in stands of amaranthus at Scissors, Texas in October 2006 that they had conspicuously damaged the plants.

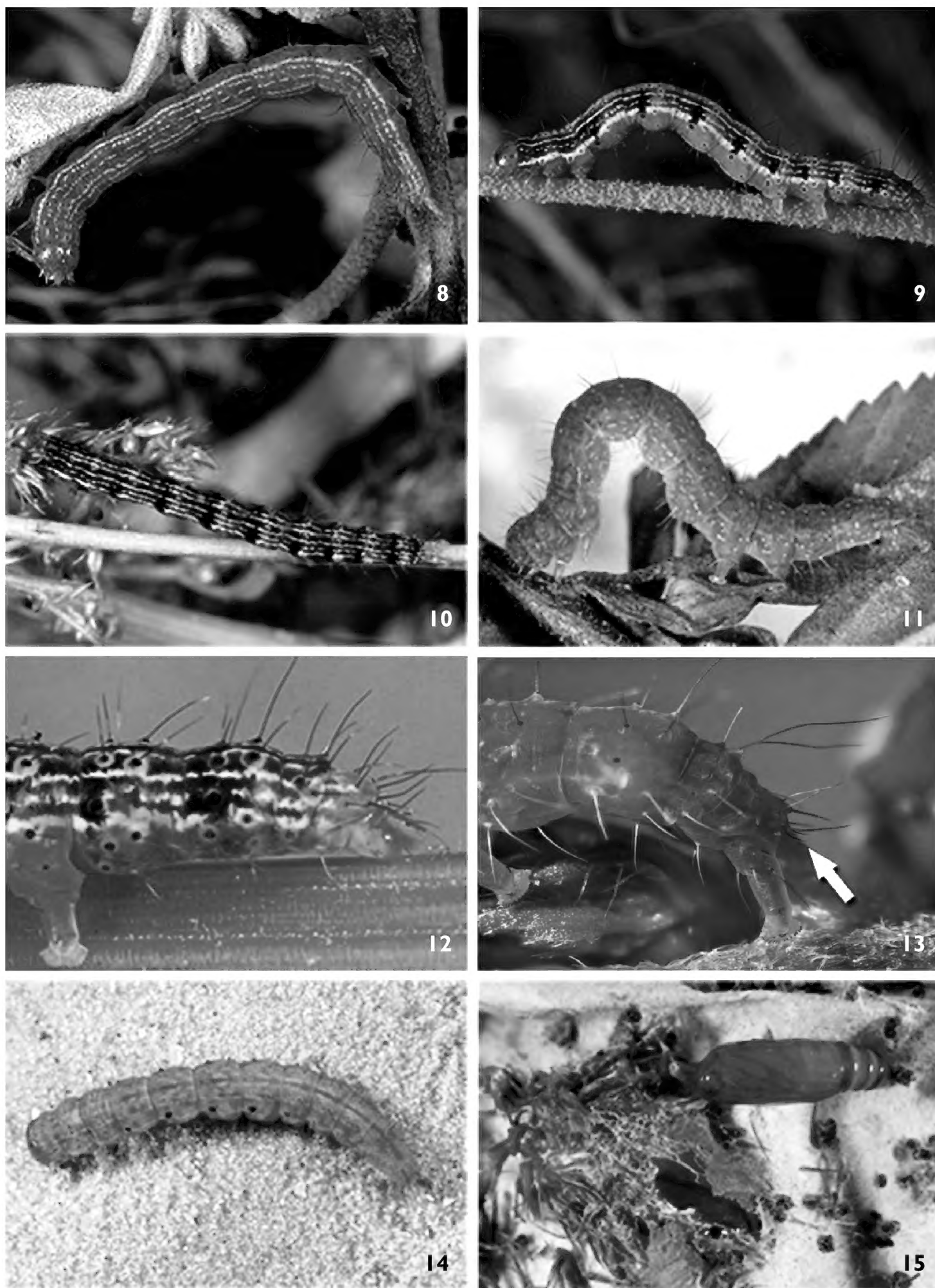
Larvae perch on the undersides of leaves and along stems and petioles. At rest, the abdominal segments are often looped upward (Fig. 9). When alarmed the larva essentially jumps from the host and continues to writhe and wriggle wildly. A QuickTime® movie of the alarm response of *Amyna* is mounted with this paper on <http://www.eeb.uconn.edu/people/wagner/>.

Prepupal larvae take on a pinkish cast (Fig. 14). Pupation occurs in a silken cocoon below (usually) or at the soil surface; sand and/or plant debris are interwoven into the cocoon wall. Adults occur throughout the growing season in southern Texas, but numbers peak in September and October during the wet season (Ed Knudson, personal communication).

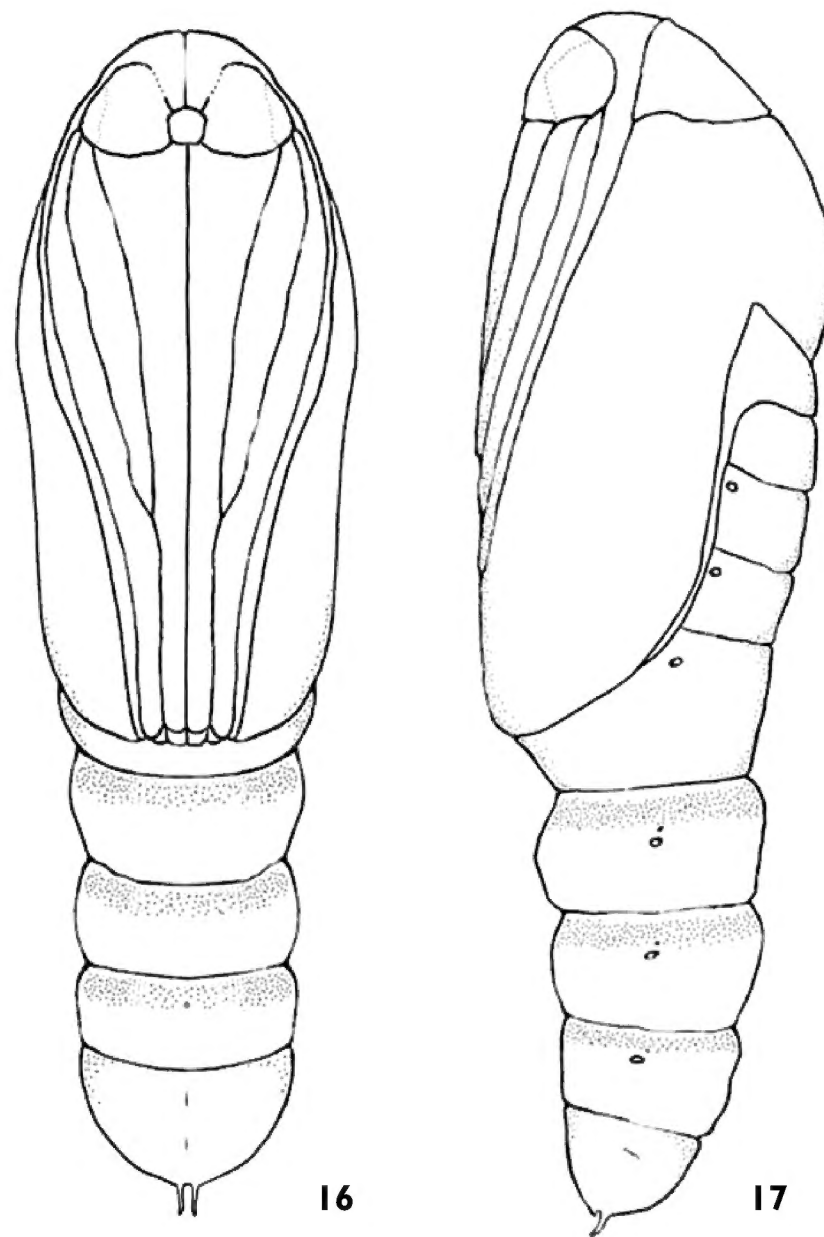
At low densities caterpillars are pale green ($n > 20$). Where we encountered caterpillars in high density and in our rearing containers, we saw higher frequencies of melanistic phenotypes. The degree of blackening varied widely, ranging from individuals with simple subdorsal-lateral patches (Fig. 9) to those that were mostly black (Fig. 10). In the field, melanized individuals (common in 2006) were visible from many meters away.

Discussion

Taxonomic matters. Crumb (1956) was so perplexed by the taxonomic affinities of *Bagisara* that he placed the genus into its own subfamily in his monograph, noting that “All of the principal characters of the larvae are primitive but the moths are rather stout and stubby winged, showing none of the superficial characters students of the moths have come to associate with lower Phalaenidae [Quadrifinae].” Kitching and Rawlins (1998) concurred with Crumb’s assessment and recognized Bagisarinae as a distinct



Figures 8–15. *Amyna axis* and *Bagisara repanda* last instar and pupa. **8–10** *Amyna axis* **11** *Bagisara repanda* **12** *Amyna axis* caudal segments **13** *Bagisara repanda* caudal segments; with D2 setae indicated by white arrow **14** *Amyna axis* prepupa **15** *Amyna axis* pupae and cocoon.



Figures 16–17. Pupa of *Amyna axis*. **16** ventral **17** lateral.

subfamily of trifine noctuids. The subfamily's position remains essentially unchanged in Fibiger and Lafontaine (2005), Lafontaine and Fibiger (2006), and Lafontaine and Schmidt (in press), i.e., at the base of the trifines in the vicinity Plusiinae, Eustrotiinae, and Acontiinae.

Character evidence that argues for a close phylogenetic association between *Amyna* and *Bagisara* include the (1) elongate body, with shallow incisures between segments and tapered caudal segments (Figs 8–13); (2) prolegs absent or greatly reduced on A3 and A4; (3) at least some crochets with a subapical tooth (Fig. 7); (4) spiracle on A1 $1.2 \times$ that on A2 and that on A8 $2 \times$ height of spiracle on A7, yellowish; (5) spiracles on A2–A4 shifted dorsad; (6) L3 shifted anteriorad below L2 on A7; (7) setae long, especially those on T1 and A8–A10 (Figs 12, 13); (8) SD1 on A10 elongate and thickened; (9) long labial palpus, $>3 \times$ basal diameter; and (10) pink to reddish prepupae (Fig. 14). Behavioral characters suggesting that *Amyna* is a bagisarine include (11) a resting posture with the mid-abdomen looped above the substrate (Figs 9, 11); (12) an animated and prolonged alarm response (QuickTime® movies of the alarm response

of both genera are mounted on <http://www.eeb.uconn.edu/people/wagner/>); and (13) the flicking of the excreta. The appendiculate crochets and shared spiracular “formula” (i.e., with the height of the spiracle on A1 being $1.2 \times$ than that of those on A2–A7, that on A8 being $>2 \times$ that on A7, as well as the dorsal shift of the spiracles on segments A2–A4), strike us as compelling evidence of (recent) common ancestry, and support Holloway’s (2009) suggestion that *Amyna* should be transferred to Bagisarinae.

Conversely, numerous structural differences separate the two genera. *Bagisara* (and some *Xanthodes* Guenée) are unusual among noctuids in their possession of two SV setae on A7 (Crumb 1956, Kitching and Rawlins 1998), whereas *Amyna axis* has only a single SV seta on A7. The prolegs are represented on A3 and A4 in *Amyna axis* by minute nibs; there is no evidence of the prolegs on these segments in the *Bagisara* that we examined. The basal tooth on the crochet of *Amyna* is much less developed than that seen in *Bagisara* species, although some Indian members of the genus evidently have a well-developed subapical tooth (Gardener 1946). *Bagisara* possess short, stout, upcurved D2 setae on A10 (Fig. 13) that presumably play an important role in fecal flicking (see Weiss 2006); the D2 setae are straight and much longer in *Amyna*. *Bagisara* are dietary specialists with much of their species radiation tied to the Malvaceae, whereas *Amyna axis* has been reared from a wide array of forbs.

Like Crumb (1956), we confess to being confused about the phylogenetic affinities of the Bagisarinae. *Bagisara* was long classified in the Acontiinae (e.g., Forbes 1954, Franclemont and Todd 1983) and most recently in its own subfamily proximate to the Acontiinae and Eustrotinae (e.g., Kitching and Rawlins 1998, Fibiger and Lafontaine 2005, Lafontaine and Fibiger 2006, Holloway 2009). In form and habit, the larvae of *Bagisara* (with *Amyna*) share similarities with Acontiinae (such as *Tarachidia* Hampson): e.g., trifold venation in adults, frass flicking habit, absence of prolegs on A3 and A4, and a simple cremaster. But other features, especially in the larvae, suggest linkages with erebids and especially the genus *Anomis* Hübner (Scoliopteryginae): in both the SV group is trisetose on A1 and A2 and the crochets are appendiculate. In addition, *Anomis* and *Bagisara* are Malvaceae specialists. Still other traits are shared with the Hypeninae and Scoliopteryginae taken together: all have some reduction of the prolegs on A3 and A4, flick their frass, have a sustained thrashing response when alarmed, and a spiracular stripe that continues on to the anal plate.

Melanism matters. *Amyna axis* larvae are green at low densities in the wild, but at high larval densities — several per sq. m — we encountered larvae with varying amounts of dark pigmentation (Fig. 10). We have also induced blackening in our rearing containers; green, unmarked larvae reared together usually darken within days of their collection. Homologous green-to-black polymorphism occurs in more than two dozen other lepidopteran lineages (Wagner in prep.). In a fashion similar to that seen in *Amyna*, caterpillars of the elm spanworm, *Ennomos subsignarius* (Hübner), darken as a result of physical contact with other larvae (Drooz 1966). Green-to-black color changes have also been linked to crowding in other noctuids: e.g., *Plusia gamma* L. (Long 1953) and *Mamestra brassicae* (L.) (Goulson 1994). In addition to crowding, temperature can induce color polyphenisms in caterpillars. Arizona populations of

Battus philenor (L.) raised in high temperatures display a red (cooler core) larval phenotype, whereas those raised at lower temperatures displayed a black (warmer core) phenotype, suggesting that color changes could be a way of “beating the heat” (Nice and Fordyce 2005). The eastern black swallowtail, *Papilio polyxenes* (Stoll), has a dark, cool-season phenotype that has thermoregulatory advantages over the lighter mid-summer morphs (Hazel 2002). Long (1953) reported that the degree of blackening in *Plusia gamma* was also influenced by a caterpillar’s diet. In *Amyna*, the production of dark forms appears to be linked to crowding as such were observed at high field densities and were induced in rearing containers with multiple larvae. Whether or not the green-to-black shift in *Amyna* and other caterpillars represents an adaptive response and the selective consequences of a shift to darker coloration change remain to be elucidated.

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